

MATING BEHAVIOUR OF *LYMNAEA STAGNALIS*

Y.A. van Duivenboden and A. ter Maat¹

*Department of Biology, Free University, P.O. Box 7161,
1007 MC Amsterdam, The Netherlands*

ABSTRACT

Mating behaviour of the pond snail *Lymnaea stagnalis* was analysed in pairs of snails, reunited after a period of isolation. Apparent female behaviour is absent in this situation, whereas male behaviour is characterized by a series of consecutive acts: mounting, turning, eversion of preputium, intromission, withdrawal of preputium and moving off. Several variations on this basic pattern occur, of which sham-copulation (false coupling) and reciprocation (reversal of roles after completion of copulation) are the most remarkable ones. The same behavioural sequence appeared to be present in spontaneous matings. Prior experience is not needed for the performance of mating behaviour. The duration of the behaviour is variable. This is mainly due to the latency of intromission. The duration of intromission is fairly constant (36 ± 4 min). Inspection of the vagina of female copulants for the presence of semen revealed that 90-100% of the copulations is successful. Surgical removal of the part of the vas deferens that runs through the body wall eliminates all male copulation behaviour without affecting the ability of copulation as a female.

Key words: *Lymnaea stagnalis*, mating, male and female behaviour.

INTRODUCTION

The reproductive biology of the simultaneous hermaphrodite freshwater snail *Lymnaea stagnalis* (Linnaeus) has received much attention in recent years. The first systematic study of the relation between copulation and egg laying was conducted by Horstmann (1955). Since then, laboratory studies have been done on endocrine and neurophysiological control of ovulation and oviposition, the structure and maturation of the reproductive system, external factors affecting fecundity and related issues (for reviews see Joosse & Geraerts 1983, Geraerts & Joosse 1984). An experimental study on the fecundity of *L. stagnalis* in the field was presented by Brown (1979).

Mating in freshwater pulmonate snails—mostly simultaneous hermaphrodites—is not a necessary condition for egg-laying in many species: under conditions of isolation they may reproduce by internally self-fertilized eggs (Duncan, 1975, Geraerts & Joosse, 1984). Yet mating must be considered a major element in the reproduction tactics of these hermaphrodite animals for several reasons: (1) Studies using genetic markers dem-

onstrated that after the snails have been allowed to mate, cross-fertilized offspring is produced for some weeks by animals that have copulated as females (*Biomphalaria glabrata*: Paraense, 1956, 1959, Richards, 1970; *L. stagnalis*: Cain, 1956). (2) In *L. stagnalis*, isolated before the appearance of sexual behaviour, the onset of egg-laying is delayed by two or more weeks due to the absence of foreign semen (Van Duivenboden, 1983). (3) Once egg-laying has started, mating reduces fecundity in those lymnaeids studied (De Witt & Sloan, 1958; Van Duivenboden *et al.*, 1985).

Under laboratory conditions *L. stagnalis* commences mating activity at the age of 7-8 weeks (shell height about 18 mm). Egg-laying starts 2-3 weeks later. Once egg-laying has started, the snails show continuous mating and oviposition activity.

A general description of the copulation behaviour of *L. stagnalis* was given by Noland & Carriker (1946) and a more detailed one by Barraud (1957). The latter concluded that copulation was seldom successful. This seems to contradict the studies mentioned above, because it implies that the complex and time-consuming mating behaviour hardly

¹To whom all correspondence should be addressed.

serves any purpose in reproduction. Moreover, Barraud suggested that a systematic study of mating in this snail is hardly possible.

In recent years we studied masculinity and receptivity in *L. stagnalis* (Van Duivenboden & Ter Maat, 1985) and effects of mating on egg-laying (Van Duivenboden, 1983, Van Duivenboden *et al.*, 1985). From these studies a detailed description of the mating behaviour of *L. stagnalis* was compiled, which is presented here.

Copulation behaviour can be induced readily by reunion of snails after a period of isolation (Noland & Carriker, 1946, Rudolph, 1979a). This method was used to describe mating behaviour and the results were compared with spontaneous matings as well. The success of copulation was assessed through dissection of female copulants, immediately after mating. Finally a simple operation was performed, which completely eliminates male mating activity.

METHODS

Laboratory-bred specimens of *L. stagnalis* were used. They were raised and kept in tanks with continuous water change at a temperature of $20 \pm 1^\circ\text{C}$ (Van der Steen *et al.*, 1969). A 12/12 light/dark cycle was maintained with overhead fluorescent lighting.

Isolation was performed by placing the animals individually in perforated polyethylene jars in the tank. They were fed lettuce leaves *ad libitum* (cf. Scheerboom, 1978). After six or more days of isolation, mating was induced by housing the animals in pairs in clean jars (one pair per jar) filled with 250 ml of fresh aerated tap water in a temperature controlled room (20°C). The snails were marked with nail polish at the tip of the shells to simplify identification during observation. At first the behaviour was observed continuously to develop criteria for the analysis of behaviour. Mating behaviour in snails is very slow so two persons can observe adequately 20–25 pairs at a time by brief observations at 1 min intervals.

Spontaneous mating behaviour was observed in the 800 liter breeding tanks (with up to 700 snails per tank) in the laboratory. The animals were fed three times a week, alternatively lettuce leaves and fish food (Tetraphyll, Tetrawerke A.G.), in restricted amounts.

Female copulants were dissected immedi-

ately after copulation. The vagina normally looks flaccid and transparent and it is difficult to recognize. A copulation was considered as successful when the vagina had a white, swollen appearance (3–5 times its normal size). In those cases the otherwise transparent duct of the bursa copulatrix was also filled with white material.

The animals were anesthetized with MgCl_2 (Van Duivenboden, 1982). A small cut was made in the body wall to remove some mm of the vas deferens. The vas deferens was interrupted in either one of two places: 1) where it runs freely in the rear sinus, or 2) the part that runs through the body wall (see Fig. 5 for anatomical details). Sham-operated animals were treated like the operated ones, except for the cutting of the vas deferens. Recovery required up to 4 hours.

Frequency data were tested by means of the G-test after Williams' correction. Analyses were carried out according to Sokal & Rohlf (1981). Data were tested for normality with the method of Shapiro & Wilk (1965, 1968) and for homogeneity of variances with the F-max procedure.

The terms "male" and "female" refer to the male and female copulant, respectively.

RESULTS

Isolation-induced mating

Mating behaviour of about 750 pairs of snails was observed, in a series of fifty experiments. After an isolation period of six days or longer, 75–100% of the pairs showed mating behaviour.

Male mating behaviour stood out clearly from all other behaviours exhibited by *L. stagnalis*. A number of consecutive behavioural acts could be distinguished in all successful copulations in males, but not in females. Females seemed to behave indifferently for the greater part of the time, moving about, air breathing and feeding during all phases of copulation. Although we will therefore focus on the male, some reactions of the female will be discussed. Firstly, a description of a straightforward mating sequence, will be given. This will serve as the basic framework for the treatment of all other behaviour accompanying mating.

When the isolated snails are paired, they crawl about in a seemingly random fashion. When they meet, the prospective male

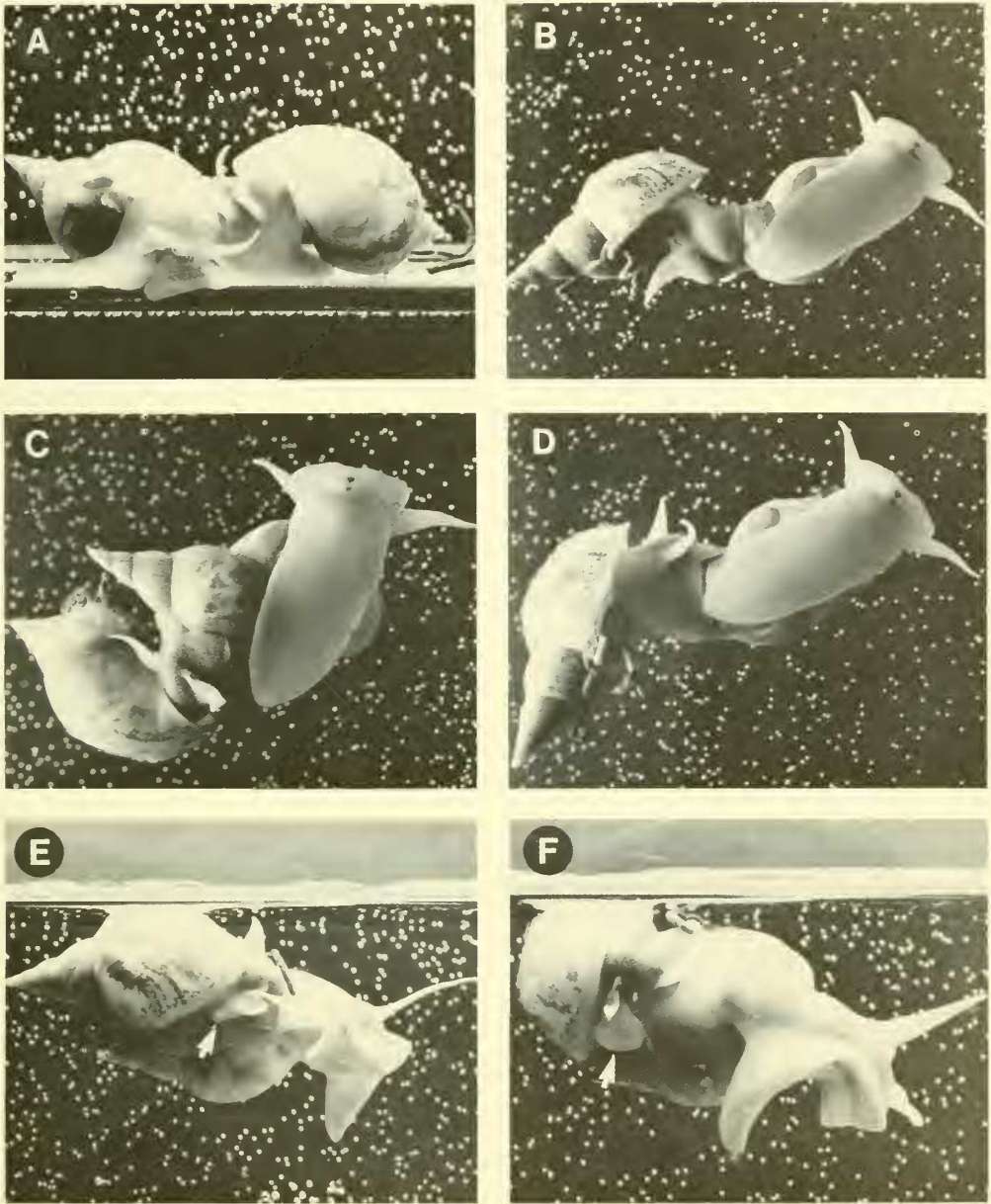


FIG. 1. A. Mounting, male at right. B, C, D. Turning, male at left. E, F. Partial eversion of preputium (arrow), male at left.

mounts the prospective female (mounting) and starts rounding the apex of her shell (turning), always in a (characteristic) counter-clockwise manner (seen from above). The male opening, near the base of the right tentacle, becomes visible as a white dot, indicating that the eversion of the preputium

had started (partial eversion of preputium) (Fig. 1A-F). When the male reaches the aperture of the shell of its partner—at the right hand side—the male comes to an almost complete stop. At this point the head/foot part shortens and gets a swollen appearance. The tentacles are shortened and drooped and the

apex of the shell is kept down. This posture is specific for mating snails. Progress is now very slow, as the right side of the foot of the male travels along the margin of the shell of the female. Meanwhile swellings and contractions occur in the partially everted preputium. Locomotion ceases altogether when the lips and tentacles of the male have passed the pneumostome of the partner. The preputium is then totally everted and the tip makes searching movements under the female's shell. A totally everted preputium is unmistakable because of its large size (length 10–20 mm, width 5–10 mm in adult snails). The actual eversion of the penis can only occasionally be observed. However, once the female gonopore of the female copulant is occupied by the tip of the preputium of the male, intromission is highly probable (see below: semen transfer). During intromission, undulations of the vas deferens are visible through the transparent wall of the preputium. The preputium is subsequently withdrawn and the male moves away. During mating the male is very firmly attached to the shell of the female: copulating animals can only forcibly be separated.

Fig. 2 summarizes the mating behavior. Thick lines in the diagram indicate the basic features of the behavioural sequence as described above. Thin lines refer to events which may complicate mating. Several mountings, whether or not followed by turning and rarely even by partial eversion of the preputium, may occur, after which the animals separate and start again. The snail which was the first to mount will generally, but

not always, become the male. Occasionally reversal of roles occurs during the mounting or turning phase. During the phase of partial eversion of preputium, role reversal is very rare and during the phase of total eversion of the preputium role reversal never occurs.

Another complication occurs when the preputium is totally everted before the male is in the right position. Two things may happen then: (1) the preputium makes some searching movements under the female's shell, is then partially withdrawn, and the male makes one or more turns followed by a second attempt, or (2) a "sham"-copulation takes place, i.e. the preputium is put under the shell of the female without subsequent intromission and ejaculation.

A sham-copulation is generally characterized by strong withdrawal of the forepart of the female, after which she relaxes again and may resume locomotion or floating, while the preputium remains in place (Fig. 3A–C). This situation continues for 15–60 min, or even for some hours. In most sham-copulations the preputium is placed between the tentacles of the female (frontally) but every position at the margin of the shell is possible. The preputium may even be inserted in the pneumostome of the partner. Sham-copulation comes to an end by partial withdrawal of the preputium. One or more turns are then made, followed by a second attempt. When the male was already in the correct position, the turns may be omitted. Up to two successive sham-copulations may precede intromission. Sham-copulations occur frequently ($\geq 50\%$ of the pairs, see Table 1).

TABLE 1. Latency from pairing and duration of intromission and the occurrence of sham-copulation in isolation-induced matings. A.: Snails with shell heights of 31–35 mm, aged 13–17 weeks. B: Snails with shell heights of 21–24 mm, aged 9 weeks.

Period of isolation (days)	N	Intromission				Sham-copulation
		Latency		Duration		
		Mean \pm SD (min)	CV ^a	Mean \pm SD (min)	CV ^a	
A. 6	20	158 \pm 46	0.29	36.4 \pm 3.2	0.09	50%
8	5	143 \pm 68	0.47	34.6 \pm 2.7	0.08	80%
11	16	135 \pm 51	0.38	36.5 \pm 4.1	0.11	94%
16	13	89 \pm 31	0.35	36.2 \pm 3.2	0.09	77%
B. 16 ^b	31	95 \pm 29	0.30	35.9 \pm 3.7	0.10	62%

^aCV = Coefficient of Variation.

^bIsolated before the appearance of sexual activity.

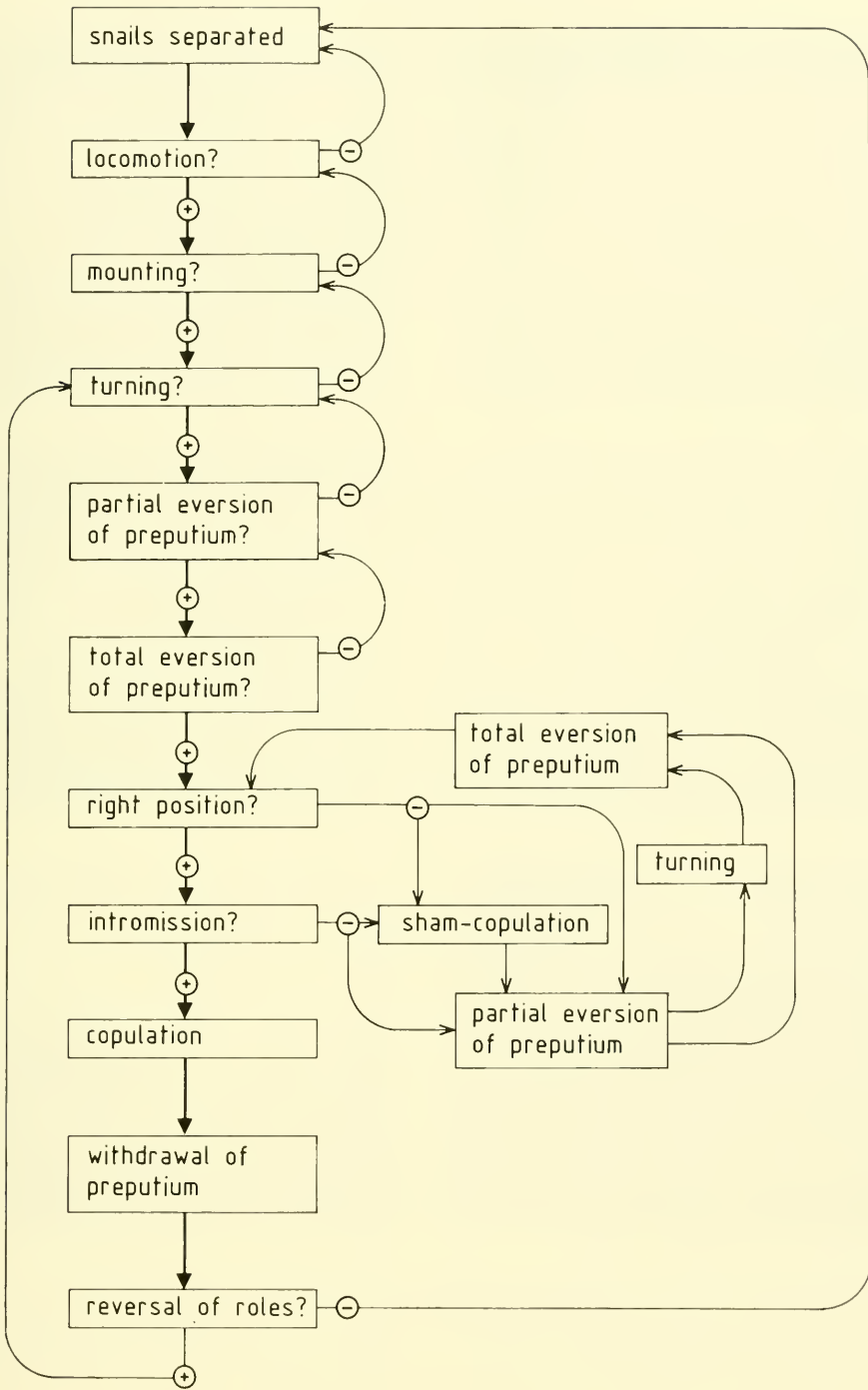


FIG. 2. Diagram of the mating behaviour of *L. stagnalis*. Thick lines refer to the basic features of male mating behaviour. Thin lines refer to behaviours which often accompany mating. See text for details.

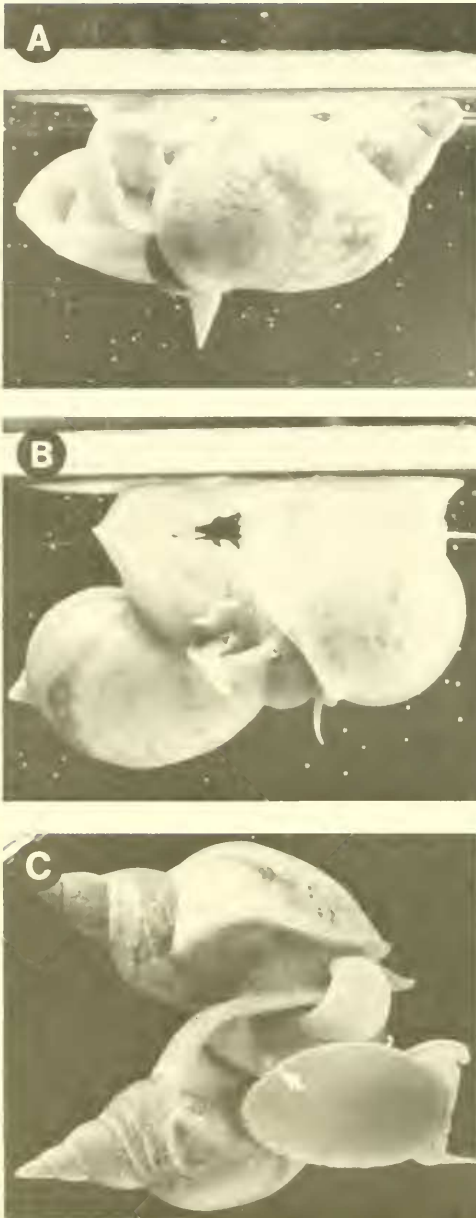


FIG. 3. A. Contorted retraction of the female (at left). B. Relaxation of the female (at left). C. Fully relaxed female resuming locomotion (lower snail), carrying the male upon her shell. Arrow: unoccupied female gonopore of the female, indicating sham-copulation.

During intromission the female may mount the shell of the male, while *in copulo*. This results in an extremely complex posture of

both snails (Fig. 4A–E). After intromission is completed in this situation, the male loses foothold and a second mating sequence with the roles reversed takes place. Reciprocal copulation contains all the basic behaviours but the introductory behaviour is shortened. When both snails have acted as male and female in turn (Fig. 4F), no mating attempt occurs for at least 4 hrs. Once total eversion of the preputium is observed, completion of the mating sequence will be the rule, with a few exceptions. Occasionally a female half-way through the copulation sequence climbs some cm above water level and falls down. Then the copulants may become separated. The other exception may occur when the female start ovipositing. Then in some cases the preputium is totally withdrawn and the male moves away. When the deposition of the egg mass is finished, the whole mating sequence may restart, sometimes with the former female in the male role. In other cases the male waits with its preputium partially everted, while attached to the female, until the egg mass is deposited. Then a second, generally successful, attempt follows.

During the entire mating sequence, the partners may have mutual mouth contact from time to time. The longer the periods of isolation, the more frequently this behaviour occurs.

The time relationships between the main male behaviours—turning, partial and total eversion of preputium and intromission—appeared to be variable. In a given experiment, e.g. in some pairs the sequence of mounting, turning and partial eversion of the preputium may take only a few minutes and the total eversion of preputium may occur as much as 90 min later. In other pairs the time between turning and partial eversion of the preputium may take more than an hour whereas it is followed immediately by total eversion of preputium and intromission. In Table I the analysis of the latency from pairing to intromission and the duration of intromission is summarized for four experiments. The coefficient of variation (CV) of the latency of intromission (0.29–0.47) appeared to be much higher than that of its duration (0.08–0.11). A clear trend towards decreasing mean latencies with increasing periods of isolation occurs ($P < 0.001$, linear regression). No such trend could be found in the mean duration of intromission, which is fairly constant (36 ± 4 min).

The latency and the duration of intromis-

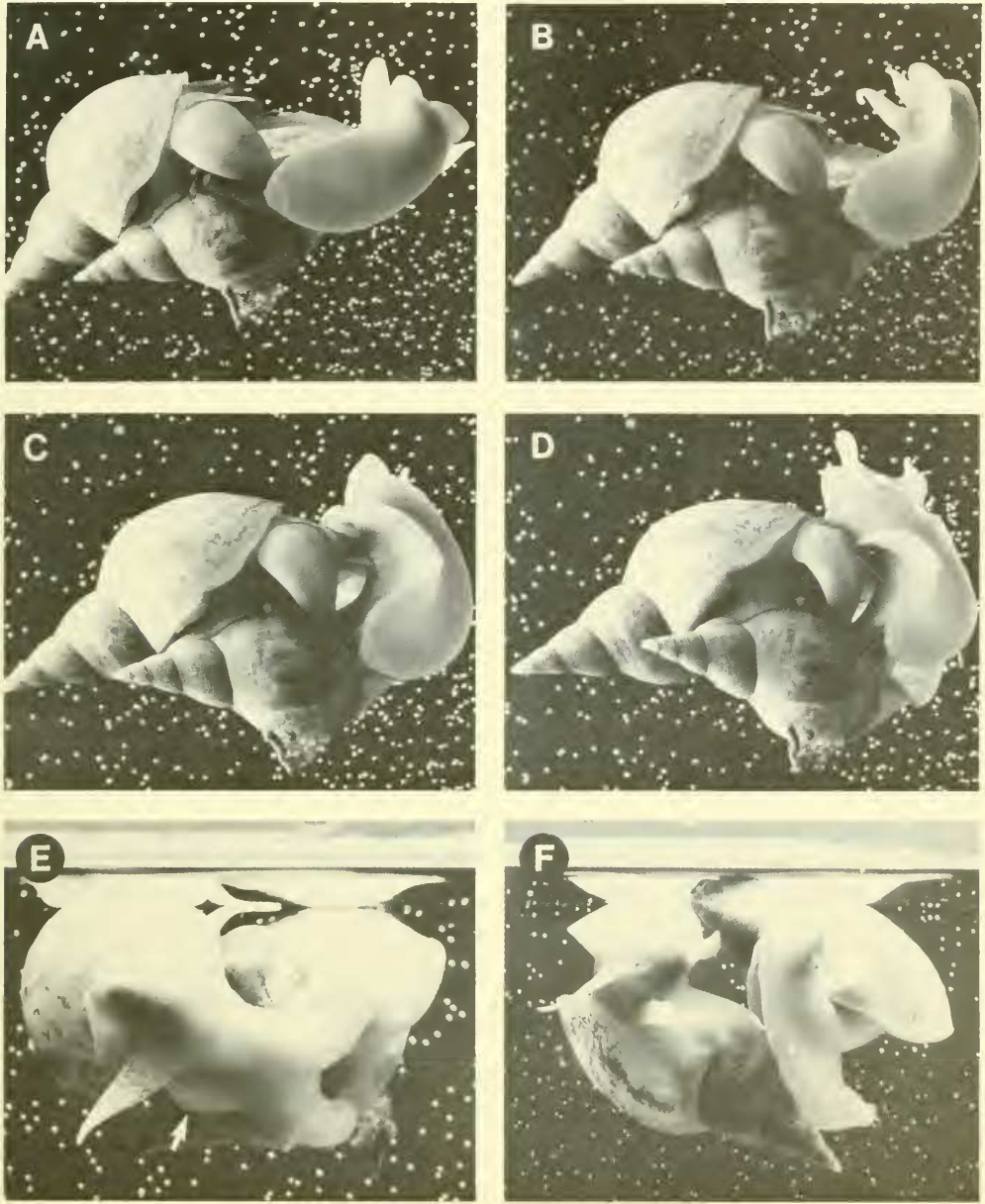


FIG. 4. A. Intromission, male at left. B, C, D. Female (at right) climbing upon the shell of the male while *in copulo*. E. Complex posture of the two snails. From left to right: shell of the male, head/foot of the female, head/foot of the male, shell of the female. Arrow: preputium of the male. F. Separation of the snails after both have acted as male and female in turn.

TABLE 2. Presence of semen in the vagina of female copulants after isolation-induced and spontaneous mating.

	Shell height (mm)	N semen/ N observed	Percentage
Isolation-induced	≥26	59/60 ^a	98%
Spontaneous	18-22	28/32	88%
	23-27	23/25	92%
	≥28	23/24	96%

^aPooled observations from four experiments.

TABLE 3. Intromission in isolation-induced mating of Operated (partial removal of vas deferens, body wall) and Sham-operated snails. Reciprocal intromission was excluded.

snails	Intromission		Test on homogeneity	G	df	P
	+	-				
Op × Op	0	6	Overall	9.889	2	0.005 < P < 0.01
Op × SH	3	3	Op × Op/Op × Sh, Sh × Sh	8.604	1	0.001 < P < 0.005
Sh × Sh	5	1	Op × Sh × Sh	1.354	1	> 0.4 NS

sion of inexperienced snails (Table 1B) agrees very well with that of experienced snails after 16 days of isolation (Table 1A, fourth row). Intromission occurred in 13 out of 18 pairs in the experienced snails (72%) and in 31 out of 40 pairs in the inexperienced snails (77.5%). These data indicate that copulation ability does not depend on prior experience.

Spontaneous mating

For comparison spontaneous matings in groups of snails were observed. The behaviours described for the isolation-induced matings were also present in spontaneous ones. Reversal of roles after copulation was not observed in these groups, but sometimes a copulating female mounted a third snail and started male behaviour, with the copulating male passively on its shell. Occasionally chains of three snails *in copulo* were encountered, the upper one acting as a male, the middle one acting as its female partner and as a male copulant for the undermost snail. The undermost female sometimes mounted a fourth snail, but chains of more than three animals *in copulo* were not observed.

Sham-copulations were frequently encountered in grouped snails. As in the isolation-induced matings a sham-copulation was generally followed by intromission. This fact was

used to determine the duration of intromission in spontaneous copulations. Twelve sham-copulating pairs (shell height 28–33 mm) were followed until the completion of copulation. The mean duration of intromission was 35.8 ± 3.5 min. This value corresponds with that of the isolation-induced copulations (Table 1).

Semen transfer

The data in Table 2 show that in most cases transfer of semen took place. Apparently, larger snails have more success but the relevant differences are not significant ($P > 0.5$, G-test for homogeneity).

Whether the presence of ejaculate in the vagina prevents insemination by a second male, as in some insects (Parker, 1970), was investigated in the following experiment.

Twenty-four snails were paired (12 pairs) after 8 days of isolation. In 10 pairs intromission took place. Immediately after the first mating, the snails were separated to prevent reciprocal copulation. Subsequently 5 pairs consisting of former females were formed and behaviour was observed during the next 150 min. In four pairs intromission took place with durations of 33, 34, 36 and 39 min, respectively. Afterwards the vagina of the ten snails was inspected for semen. In all of them semen was present and in three of the four snails, that had acted twice as a female, the

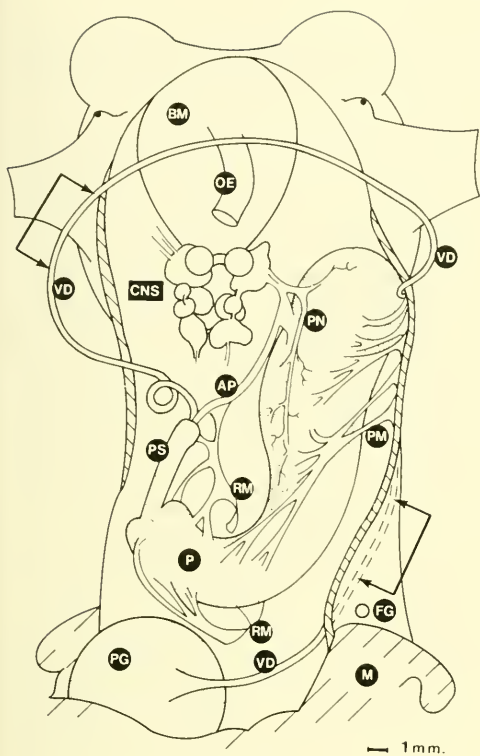


FIG. 5. Dorsal view of the internal organization of the head/foot part of *L. stagnalis*. Arrows refer to the parts of the vas deferens that were surgically removed. AP = artery of the penis, BM = buccal mass, CNS = central nervous system, FG = female gonopore, M = mantle, OE = oesophagus, P = preputium, PG = prostate gland, PM = protractor muscles, PN = penis nerve, PS = penis sheath, RM = retractor muscles, VD = vas deferens.

amount was much larger than ever observed before, indicating that insemination had occurred by the second male. Thus the presence of semen in the vagina does not prevent intromission. Moreover insemination by a second male is highly probable.

Elimination of male mating activity

In pilot studies attempts were made to block semen transfer by surgical removal of different parts of the vas deferens (see Fig. 5 for anatomical details).

Of 16 animals with part of the vas deferens in the head sinus removed, 8 animals survived (50%). Eight days after the operation the animals showed male behaviour (i.e. par-

tial eversion of preputium), but total eversion of preputium and intromission were impaired. Dissection revealed that the loose ends of the vas deferens had grown into the body wall, making total eversion of preputium and intromission impossible.

Ten animals with the part of the vas deferens that runs through the body wall removed, all survived (100%). Animals operated this way not only lacked the possibility of semen transfer, but also failed to show mating activity, even when they were paired after three weeks of isolation. Therefore a more thorough study was made of the effects of this lesion.

The operation was done in 18 snails (Op) and 18 snails were sham-operated (Sh). Afterwards the snails were kept in isolation during a period of 8 days. They were divided in three experimental groups: 6 pairs of operated snails (Op × Op), 6 pairs consisting of an operated and a sham-operated snail (Op × Sh) and 6 pairs of sham-operated snails (Sh × Sh). The behaviour of the pairs was observed during 330 min.

In all pairs mounting was observed. No differences in the latency of mounting between the groups were observed (one-way ANOVA, on log transformed data, $0.10 < P < 0.25$, NS). In the Op × Sh-group all first mountings were made by the sham-operated snail. In the Op × Op-group no copulation behaviour followed, except rarely an incomplete turn. In all pairs of the other groups male behaviour was exhibited, but only by the sham-operated snails. The number of intromissions in the groups with at least one sham-operated snail in the pairs was significantly higher than that in the Op × Op-group (Table 3).

The operated snails did not initiate mating behaviour, but it is conceivable that copulation as a female could induce male behaviour in these snails (cf. *Stagnicola elodes*, Rudolph, 1979a). This hypothesis was rejected by the total absence of any sign of reciprocal behaviour in the operated snails after copulation as a female, whereas the sham-operated snails all exhibited male behaviour after copulation as a female (G-test $P < 0.05$). In all females—operated or sham-operated—semen was present in the vagina.

Removal of part of the vas deferens that runs through the body wall clearly eliminates all male behaviour, but it does not impair the ability to copulate as a female.

DISCUSSION

The mating behaviour of *L. stagnalis* is unilateral: one snail acts as the male and the other as the female. This is the general way of mating in lymnaeid snails (*L. peregra*: Diver *et al.*, 1925; *L. tomentosa*: Boray, 1964; *S. elodes*: Rudolph, 1979a; *L. truncatula*: Smith, 1981).

Characteristic female mating behaviour is absent, whereas male mating behaviour is clear and unmistakable. The basic features are: mounting, turning, eversion of preputium, intromission, withdrawal of preputium and moving away. Several variations are present of which sham-copulation and reciprocation are the most remarkable ones.

In many respects our description of the mating behaviour of *L. stagnalis* is in agreement with the results of Barraud (1957), but there are some contradictions. Firstly, Barraud suggested that, in addition to the copulatory position that we described (position 1, Barraud), intromission is possible from a frontal position (position 2, Barraud). Secondly, at the moment of actual penetration, we did not observe any reaction of the female, whereas he described a contorted retraction of the whole forepart of the female's body at intromission, like we observed in sham-copulations. Thirdly the duration of intromission was 36 ± 4 min in our experiments, whereas Barraud reported durations of a few min to twelve hours. A fourth difference relates to the success of copulation, which was nearly 100% in our study and low in his. All these differences share a common cause: Barraud did not make a clear distinction between sham-copulation and real copulation. A sham-copulation does resemble a real copulation, but an experienced observer is able to distinguish the two by the characteristic female reaction in sham-copulation. Moreover, occupation of the vagina can in most cases—e.g. with the aid of a mirror—be observed. The high incidence of sham-copulation probably explains all contradictions between our observations and those of Barraud.

Reciprocal copulation behaviour was described extensively for *S. elodes* (Rudolph, 1979a). The readiness to exhibit male behaviour, induced in female copulants, lasts 30–60 min in this snail. When stimulated females are transferred to a third snail during this period, they behave as males. In groups the induced male behaviour of female

copulants is probably directed towards a third snail rather than to the partner, since chain copulations but no reversal of roles were observed in groups of snails.

Mouth contact was sometimes encountered during the mating sequence of *L. stagnalis*. It is a common feature of mating behaviour in snails. In helicids (terrestrial pulmonates) courtship commences with mouth to mouth contact (Lind, 1976) and it is a characteristic part of the mating behaviour of the opisthobranch *Aeolidia papillosa* (Longley & Longley, 1984). In *L. stagnalis* it is not an integrated part of mating behaviour.

The mating behaviour of *L. stagnalis* can be broken off in the first stages of the sequence (mounting, turning and occasionally partial eversion of preputium). Once the preputium is totally everted, the sequence will come to completion, although this may take hours.

Mating capability in *L. stagnalis* depends on maturation only, not on prior experience (Table 1). This has been found earlier in *Biomphalaria globosus* (Rudolph, 1983). The duration of intromission was found to be independent of the experience or the period of isolation of the snails. Probably the duration is determined by neuronal timing circuitry, as is assumed to be the case in *A. papillosa* (Longley & Longley, 1984).

After copulation as a female, the readiness to mate as a male as well as a female remains the same. The readiness to mate disappears when both snails have acted as male and female in turn. These observations as well as the decrease in the latency of intromission with increasing period of isolation is in accordance with our model of masculinity and receptivity (Van Duivenboden & Ter Maat, 1985).

Extirpation of the part of the vas deferens that runs through the body wall eliminates all male behaviour in *L. stagnalis*. As yet it is not clear whether this is due to neurological, endocrinological or mechanical blockade. Little is known of mechanisms controlling mating behaviour in other snails. Jeppesen (1976) extirpated various parts of the reproductive system of *Helix pomatia*, but the initiation and the sequence of mating behaviour were not affected. He concluded that mating behaviour is controlled by the central nervous system. The cycle of the mating behaviour in helicids (Lind, 1976, Jeppesen, 1976) seems to depend partly on copulation itself, as in *L. stagnalis* (Van Duivenboden & Ter Maat, 1985) and on mechanical effects of dart-

shooting, a behaviour not present in *L. stagnalis*. The organization of the male and the female reproductive system of *L. stagnalis* (diaulic) is different from that in *Helix* (monaulic) (Visser, 1977, 1981, Geraerts & Joosse, 1984, Tompa, 1984). Therefore a comparison of the extirpations carried out by Jeppesen in *Helix pomatia* with the lesion carried out in this study in *L. stagnalis* is not in order.

In almost all cases, copulation appeared to be successful, i.e. semen could be observed in the vagina of the female. Similar results were found for *S. elodes* (Rudolph, 1979a) and for *B. globosus* (Rudolph, 1979b, 1983). As in *Bulinus*, the presence of semen in the female tract does not prevent intromission and insemination by a second male.

ACKNOWLEDGEMENTS

The authors thank Dr. W.J. van der Steen and Prof. Dr. T.A. de Vlioger for critical reading of the manuscript, Anton Pieneman for technical assistance, photography and preparing of the figures, and Thea Laan for typing the manuscript.

REFERENCES CITED

- BARRAUD, E. M., 1957, The copulatory behaviour of the freshwater snail (*Lymnaea stagnalis* L.). *British Journal of Animal Behaviour*, 5: 55–59.
- BORAY, J. C., 1964, Studies on the ecology of *Lymnaea tomentosa*, the intermediate host of *Fasciola hepatica*. *Australian Journal of Zoology*, 12: 231–237.
- BROWN, K. M., 1979, Effects of experimental manipulations on the life history pattern of *Lymnaea stagnalis appressa* Say (Pulmonata: Lymnaeidae). *Hydrobiologia*, 65: 165–176.
- CAIN, G. L., 1956, Studies on cross-fertilization and self-fertilization in *Lymnaea stagnalis appressa* Say. *Biological Bulletin* (Marine Biological Laboratory, Woods Hole), 111: 45–52.
- DIVER, C., BOYCOTT, A. E. & GARSTANG, S., 1925, The inheritance of inverse symmetry in *Limnaea peregra*. *Journal of Genetics*, 15: 113–200.
- DUIVENBODEN, Y. A. VAN, 1982, Non-ocular photoreceptors and photo-orientation in the pond snail *Lymnaea stagnalis* (L.). *Journal of Comparative Physiology*, 149: 363–368.
- DUIVENBODEN, Y. A. VAN, 1983, Transfer of semen accelerates the onset of egg-laying in female copulants of the hermaphrodite freshwater snail, *Lymnaea stagnalis*. *International Journal of Invertebrate Reproduction*, 6: 249–257.
- DUIVENBODEN, Y. A. VAN & TER MAAT, A., 1985, Masculinity and receptivity in the hermaphrodite pond snail *Lymnaea stagnalis*. *Animal Behaviour*, 33: 885–891.
- DUIVENBODEN, Y. A. VAN, PIENEMAN, A. W. & TER MAAT, A., 1985, Multiple mating suppresses fecundity in the hermaphrodite freshwater snail *Lymnaea stagnalis*: a laboratory study. *Animal Behaviour*, 33: 1184–1191.
- DUNCAN, C. J., 1975, Reproduction. In *Pulmonates*, FRETTER, V. & PEAKE, J., eds., vol. 1, Academic Press, London, pp. 309–365.
- GERAERTS, W. P. M. & JOOSSE, J., 1984, Freshwater snails (Basommatophora). In *The Mollusca*, WILBUR, K. M., ed., vol. 7, Reproduction, Academic Press, New York, pp. 142–208.
- HORSTMANN, H. J., 1955, Untersuchungen zur Physiologie der Begattung und Befruchtung der Schlammschnecke *Lymnaea stagnalis* L. *Zeitschrift für Morphologie und Ökologie der Tiere*, 44: 222–268.
- JEPPESEN, L. L., 1976, The control of mating behaviour in *Helix pomatia* L. (Gastropoda: Pulmonata). *Animal Behaviour*, 24: 275–290.
- JOOSSE, J. & GERAERTS, W. P. M., 1983, Endocrinology. In *The Mollusca*, WILBUR, K. M. & SALEUDDIN, A. S. M., eds., vol. 4, part 1, Physiology, Academic Press, New York, pp. 317–406.
- LIND, H., 1976, Causal and functional organization of the mating behaviour sequence in *Helix pomatia* (Pulmonata, Gastropoda). *Behaviour*, 59: 162–202.
- LONGLEY, A. J. & LONGLEY, R. D., 1984, Mating in the gastropod mollusc *Aeolidia papillosa*: behaviour and anatomy. *Canadian Journal of Zoology*, 62: 8–14.
- NOLAND, L. E. & CARRIKER, M. R., 1946, Observations on the biology of the snail *Lymnaea stagnalis* during twenty generations in laboratory culture. *American Midland Naturalist*, 36: 467–493.
- PARAENSE, W. L., 1956, A genetic approach to the systematics of planorbid molluscs. *Evolution*, 10: 403–407.
- PARAENSE, W. L., 1959, One-sided reproductive isolation between geographically remote populations of a planorbid snail. *American Naturalist*, 93: 93–101.
- PARKER, G. A., 1970, Sperm competition and its evolutionary consequences in the insects. *Biological Review*, 45: 525–567.
- RICHARDS, C.S., 1970, Genetics of a molluscan vector of Schistosomiasis. *Nature*, 227: 806–810.
- RUDOLPH, P. H., 1979a, The strategy of copulation of *Stagnicola elodes* (Say) (Basommatophora: Lymnaeidae). *Malacologia*, 18: 381–389.
- RUDOLPH, P.H., 1979b, An analysis of copulation

- in *Bulinus (Physopsis) globosus* (Gastropoda: Planorbidae). *Malacologia*, 19: 147–155.
- RUDOLPH, P.H., 1983, Copulatory activity and sperm production in *Bulinus (Physopsis) globosus* (Gastropoda: Planorbidae). *Journal of Molluscan Studies*, 49: 125–132.
- SCHEERBOOM, J. E. M., 1978, The influence of food quantity and food quality on assimilation, body growth and egg-production in the pond snail *Lymnaea stagnalis* (L.) with particular reference to the haemolymph-glucose concentration. *Proceedings of the Koninklijke Nederlandse Akademie van Wetenschappen* (C), 81: 184–187.
- SHAPIRO, S. S. & WILK, M. B., 1965, An analysis of variances test for normality. *Biometrika*, 62: 591–611.
- SHAPIRO, S. S. & WILK, M. B., 1968, Approximations for the null distribution of the W statistic. *Technometrics*, 10: 861–866.
- SMITH, G., 1981, Copulation and oviposition in *Lymnaea truncatula* (Müller). *Journal of Molluscan Studies*, 47: 108–111.
- SOKAL, R. R. & ROHLF, F. J., 1981, *Biometry*. Freeman, San Francisco.
- STEEN, W. J. VAN DER, HOVEN, N.P. VAN DEN & JAGER, J. C., 1969, A method for breeding and studying freshwater snails under continuous water change, with some remarks on growth and reproduction in *Lymnaea stagnalis* (L.). *Netherlands Journal of Zoology*, 19: 403–468.
- TOMPA, A. S., 1984, Land snails (Stylommatophora) I. In *The Mollusca*, WILBUR, K. M., ed., vol. 7. Reproduction. Academic Press, London, pp. 47–140.
- VISSER, M., 1977, The morphology and significance of the spermoviduct and prostate in the evolution of the reproductive system of the Pulmonata. *Zoologica Scripta*, 6: 43–54.
- VISSER, M., 1981, Monaully versus diauly as the original condition of the reproductive system of Pulmonata and its bearing on the interpretation of the terminal ducts. *Zeitschrift für zoologische Systematik und Evolutionsforschung*, 19: 59–68.
- WITT, R. M. DE & SLOAN, W. C., 1958, The innate capacity for increase in numbers in the pulmonate snail *Lymnaea columella*. *Transactions of American Microscopical Society*, 77: 290–294.

Revised Ms. accepted 26 June 1986